# Genetic diversity of PMCV in Atlantic salmon in Ireland.

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### Cardiomyopathy Syndrome (CMS)

- CMS is characterized by inflammation of the heart in Atlantic Salmon and usually affects adult fish in their second year at sea.
- Originally identified in Norway in 1985.
- First reported CMS outbreak in marine farm in 2012 in Ireland.
- The Piscine Myocarditis virus (PMCV) was discovered and linked to the disease in 2010.





Rodger et al. (2014)



#### Piscine Myocarditis virus.

- Double stranded RNA virus in Totiviridae family.
- 6,688bp long, 3 open reading frames (ORFs).
- Seems to be present in most samples, regardless of whether the sample has CMS or not





### Irish PMCV Phylogeny

- Concatenated (1367bp) ORF1 and ORF3 amplicons.
- No clear geographic variation.
- Less variation than Norwegian samples.



Tighe et al. (2019) Journal of fish diseases. In press



#### Conclusions

- PMCV shows low genetic diversity in Ireland.
- Recent arrival from Southern Norway.
- Uniquely Irish amino acid substitutions in ORF3 (and ORFI).
- New substitution has arisen since 2012.
- No link between strains and CMS.
- Virus is becoming less virulent either virus is evolving or fish are.



#### Viral Whole Genome Sequencing

• Use MinION nanopore sequencer with long range PCR.







#### Nanopore Sequencing

- Allows for much longer reads Record at 2 million bp in one read.
- Individual reads diversity within samples.
- Cheap.





#### Salmonid alphavirus

- Collaborative project with University of Aberdeen/Edinburgh.
- Six recognized subtypes of the disease.
- New Irish isolate found in wrasse in 2017.
- Long range PCR + MinION







#### Host species

- 🐢 : Ballan wrasse
- < : Rainbow trout

- 🛶 : Arctic char
- 🛻 : Common dab
- : European plaice

Tighe et al. (2019) In preparation



#### Conclusions

- SAV strain is distinct enough to be new subtype: SAV7
- Using partitioned analysis showed SAV7 split from SAV6 ~400 years ago, as opposed to ~200 years ago using unpartitioned analysis.
- Make the case for salmonid alphavirus being renamed to piscine alphavirus.



#### **PMCV** whole genome sequencing

- MinION and Illumina.
- Long range PCR of genome.
- First genome generated (similar to Norway)
- Will allow for clearer picture of the phylogeny of PMCV in Ireland.
- Genome Wide Association Study (GWAS) of virus will be used to find any mutations linked to CMS.
- Diversity of viral cloud within fish will also be examined.



#### **PMCV** whole genome sequencing

- 99% similar to Norwegian strain (35 SNPs).
- I5 SNPs in ORFI
- I5 SNPs in ORF2
- 7 SNPs in ORF3
- Commonly used amplicon for ORFI only covers 2 SNPs, vs 6 SNPs for ORF3





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